Glossary[1]

Acceptance criteria[2]: Minimum standards for the performance of experimental controls and reference standards. All acceptance criteria must be met for an experiment to be considered valid.

Accuracy[2]: (a) The closeness of agreement between a test method result and an accepted reference value. (b) The proportion : of correct outcomes of a test method. It is a measure of test method performance.

Activation (of genes): The interaction of specific molecules or molecular complexes with specific genes to initiate their expression (transcription)

Adenosine triphosphate (ATP): A nucleotide involved in energy metabolism and required for RNA synthesis; it occurs in all cells and is used to store energy in the form of high-energy phosphate bonds.

Agonist: A substance that produces a response, e.g., transcription, when it binds to a specific receptor.

Androgen: A class of steroid hormone, which includes testosterone and 5α-dihydrotestosterone, responsible for the development and maintenance of the male reproductive system.

Androgen receptor: The receptor to which androgens bind.

Antagonist: A substance that inhibits a response, e.g., transcription, when it binds to a specific receptor.

Assay[2]: The experimental system used. Often used interchangeably with "test" and "test method".

BG-1: The BG-1Luc4E2 cell line was derived from BG-1 immortalized adenocarcinoma cells that endogenously express estrogen receptor and have been have been stably transfected with the plasmid pGudLuc7.ERE. This plasmid contains four copies of a synthetic oligonucleotide containing the estrogen response element upstream of the mouse mammary tumor viral (MMTV) promoter and the firefly luciferase gene.

Cell density: The density of cells growing in a monolayer in a single well of a tissue culture plate.

Cell morphology: The shape and appearance of cells grown in a monolayer in a single well of a tissue culture plate. Cells that are dying often exhibit abnormal cellular morphology.

Charcoal/dextrantreatment: Treatment of serum used in cell culture. Treatment with charcoal/dextran (often referred to as "stripping") removes endogenous hormones and hormone-binding proteins.

Culture medium: An aqueous solution containing vitamins, minerals and growth factors to support the growth of cells in culture.

[1] The definitions in this Glossary are restricted to their uses with respect to endocrine mechanisms and actions.
[2] Definition used by the Interagency Coordinating Committee on the Validation of Alternative Methods.

Coded test substances: Substances labeled by code rather than name so that they can be tested and evaluated without knowledge of their identity or anticipation of test results. Coded test substances are used to avoid intentional or unintentional bias when evaluating laboratory or test method performance.

Coefficient of variation: A statistical representation of the precision of a test. It is expressed as a percentage and is calculated as follows:

$$\left(\frac{\text{standard deviation}}{\text{mean}}\right) \times 100$$

Comprehensive test: The test performed for determination of an EC- or IC_{50} value. Compared to the range finder test the comprehensive test uses a smaller dilution factor for the concentrations tested.

Concordance[2]: The proportion of all substances tested that are correctly classified as positive or negative. It is a measure of test method performance and it is often used interchangeably with "accuracy".

Control: Substances selected for use during the research, development, protocol standardization, and validation of a proposed test method having a known response. Controls are used to evaluate the ongoing performance of a test method. All experimental controls must fall within established historical norms for an experiment to pass "acceptance criteria" and be considered valid.

Cytotoxicity: The adverse effects resulting from interference with structures and/or processes essential for cell survival, proliferation, and/or function. For most substances, toxicity is a consequence of non-specific alternations in "basal cell functions" (i.e., via mitochondria, plasma membrane integrity, etc.).

Dextran: A viscous or semi-viscous polymer of glucose.

EC_{50}: The half maximal effective concentration of a test substance.

EDSP: The U.S. EPA Endocrine Disruptor Screening Program.

EDSTAC: The U.S. Endocrine Disruptor Screening and Testing Advisory Committee.

EDWG: The ICCVAM Endocrine Disruptor Working Group, a group comprised of knowledgeable scientists from participating ICCVAM agencies.

Endocrine: Of or relating to the endocrine system, endocrine glands, or hormones.

Endocrine disruptor: Substances that interact with the endocrine system to alter normal functioning. Endocrine disruptors may act directly by activating or inhibiting a receptor, altering hormone biosynthesis or transport, or altering hormone metabolism.

Endocrine system: Comprises the glands, located throughout the body that secrete hormones, the hormones that are secreted, and the receptors that recognize and respond to the hormones.

Endpoint: The biological process, response, or effect assessed by a test method.

Essential test method components[2]: Structural, functional, and procedural elements of a validated test method that should be included in the protocol of a mechanistically and functionally similar proposed test method. These components include unique characteristics of the test method, critical procedural details, and quality control measures. Adherence to essential test method components is necessary when the acceptability of a proposed test method is being evaluated based on performance standards derived from a mechanistically and functionally similar validated test method.

False negative[2]: An active substance incorrectly identified as negative by a test.

False negative rate[2]: The proportion of all positive (active) substances falsely identified as negative. A measure of test method performance.

False positive[2]: An inactive substance incorrectly identified as positive by a test.

False positive rate[2]: The proportion of all negative (inactive) substances falsely identified as positive. A measure of test method performance.

Fluorescence: The emission of radiation, especially of visible light.

FR: The U.S. Federal Register. The Federal Register is the official daily publication for rules, proposed rules, and notices of U.S. Federal agencies and organizations.

Guidance document: ICCVAM Evaluation of Test Methods for Detecting Potential Endocrine Disruptors: Estrogen Receptor and Androgen Receptor Binding and Transcriptional Activation Assays (ICCVAM 2003).

Hill function: A four parameter logistic mathematical model relating the concentration of the test substance to the response (typically following a sigmoidal shape).

$$Y = \text{Bottom} + \frac{\text{Top} - \text{Bottom}}{1 + 10^{(\log EC_{50} - \log X)\text{HillSlope}}}$$

where Y = response (i.e., luciferase activity), X is the substance concentration producing the response, Bottom is the minimum response, Top I the maximum response, EC_{50} is the substance concentration at the response midway between Top and Bottom, and HillSlope describes the slope of the curve.

IC_{50}: The half maximal inhibitory concentration of a test substance.

Interlaboratory reproducibility[2]: A measure of whether different qualified laboratories using the same protocol and test substances can produce qualitatively and quantitatively similar results. Interlaboratory

reproducibility is determined during the validation process and indicates the extent to which a test method can be transferred successfully among laboratories.

Intralaboratory repeatability[2]: The closeness of agreement between test results obtained within a single laboratory when the procedure is performed on the same substance under identical conditions within a given time period.

Intralaboratory reproducibility[2]: The first stage of validation; a determination of whether qualified people within the same laboratory can successfully replicate results using a specific test protocol at different times.

***In vitro*:** Literally, in glass. Refers to assays that are carried out in an artificial system (e.g., in a test tube or Petri dish), and typically use single-cell organisms, cultured cells, cell-free extracts, or purified cellular components.

***In vivo*:** In the living organism. Refers to assays performed in multi-cellular organisms.

Luciferase: An enzyme present in the cells of some bioluminescent organisms that catalyzes the oxidation of luciferin and ATP to produce luminescence.

Luminescence: The emission of radiation, especially of visible light caused by chemical, or biochemical processes.

Luminometer: A device for measuring luminescence.

Negative predictivity[2]: The proportion of correct negative responses among substances testing negative.

Peer review[2]: Objective review of data, a document, or proposal, and provision of recommendations, by an expert individual or group of individuals having no conflict of interested with the outcome of the review.

Plasmid: A circle of bacterial DNA that is self-replicating. Plasmids can be artificially constructed and used as cloning vectors.

Positive predictivity[2]: The proportion of correct positive responses among substances testing positive.

ppb: Parts per billion. One part in 10^9 molecules.

ppq: Parts per quadrillion. One part in 10^{15} molecules.

Precipitate: A solid substance, often in the form of crystals, separated from a solution.

Precipitation: The act of a solid substance separating from a solution.

Protocol[2]: The precise, step-by-step description of a test, including the listing of all necessary reagents, criteria, and procedure for the valuation of the test data.

Protocol standardization: Selection of reference standards, controls, and performance standards for a protocol prior to initiation of validation efforts.

Q test: The Q test is a simple statistical test to determine if a data point that appears to be different from the rest of the data points in a set may be discarded. The Q test is

$$Q = \frac{\text{suspectedoutlier} - \text{closest value}}{\text{maximumvalue} - \text{minimumvalue}}$$

The resultant value, Q, is then compared to a table of critical values (Qc). If Q is larger than Qc, the data point is an outlier and can be discarded with 90% confidence (e.g, in a data set with values 100, 2655, and 241, the Q value is 0.95. For a set of three data points, Qc is 0.94. Q [0.95] is greater than Qc [0.94], so 2655 is an outlier and can be discarded).

Receptor: A protein of protein complex, which binds to specific molecules or the purpose of transporting them elsewhere in the cell, or for producing a chemical signal.

Receptor binding assay: An assay to measure the ability of a substances to bind to a hormone receptor protein, which is typically performed by measuring the ability of the substances to displace the bound natural hormone.

Reduction alternative: a new or modified test method that reduces the number of animals required.

Refinement alternative: a new or modified test method that refines procedures to lessen or eliminate pain or distress in animals of enhances animal well-being.

Relevance[2]: The extent to which a test method correctly predicts or measure the biological effect of interest in the species of interest. Relevance incorporates consideration of the "accuracy" or "concordance" of a test method.

Reliability[2]: A measure of the degree to which a test method can be performed reproducibly within and among laboratories over time. Reliability is assessed by calculating intra- and inter-laboratory reproducibility and intralaboratory repeatability.

Screen/screening test[2]: A rapid, simple test conducted for the purposes of a general classification of substances according to general categories of hazard. The results of a screen generally are used for preliminary decision making and to set priorities for more definitive tests.

Selection: Enrichment of stably transfected cells in tissue culture by usually by exposure to a substance that is noxious to non-transfected cells (e.g. exposure of cells to G418 kills cells that do not contain the G418 resistance vector).

Sensitivity[2]: The proportion of all positive substances that are classified correctly as positive in a test method. It is a measure of test method accuracy.

Specificity[2]: The proportion of all negative substances that are classified correctly as negative in a test method. It is a measure of test method accuracy.

Stable transfection: When DNA is transfected into cultured cells in such a way that it is stably integrated into the cells genome, resulting in the stable expression of transfected genes. Clones of stably transfected cells are selected by stable markers (e.g., resistance to G418).

Standard operating procedures (SOPs)[2]: Formal, written procedures that describe how specific laboratory operations are to be performed. These are required by GLP Guidelines.

Tier 1 assay: An assay that is a component of the EDSP screening battery of tests. Tier 1 screening will include a battery of screening assays that would identify substances that have the potential to interact with the estrogen, androgen, or thyroid hormone systems.

Tier 2 assay: An assay that is a component of the EDSP testing battery. Tier 2 tests are longer in duration than Tier 1 tests and are intended to encompass a broad range of doses, life stages, and processes.

Transactivation:

Transfection: The process by which foreign DNA is introduced into a cell to change the cell's genotype.

Transcription: Synthesis of RNA by RNA polymerases using a DNA template.

Transcriptional activation: The initiation of mRNA synthesis in response to a specific chemical signal, such as a binding of an estrogen to the estrogen receptor.

Transferability[2]: The ability of a test method or procedure to be accurately and reliably performed in different, competent laboratories.

Transient transfection: When DNA is transfected into cultured cells, but is not stably integrated into the cells genome and is only retained for two to three days.

Validated test method: An accepted test method for which validation studies have been completed to determine the accuracy and reliability of the method for a specific proposed use.

Validated method[2]: An accepted test method for which validation studies have been completed to determine the accuracy and reliability of this method for a specific proposed use.

Validation[2]: The process by which the reliability and accuracy of a procedure are established for a specific purpose.

Vector: A small segment of DNA (frequently a plasmid or viral DNA) that is used to carry a foreign gene or DNA sequence into a cell.

Weight of evidence (process)[2]: The strengths and weaknesses of a collection of information are used as the basis for a conclusion that may not be evident from the individual data.

Xenobiotic: A substance that is not produced by the organism of interest.